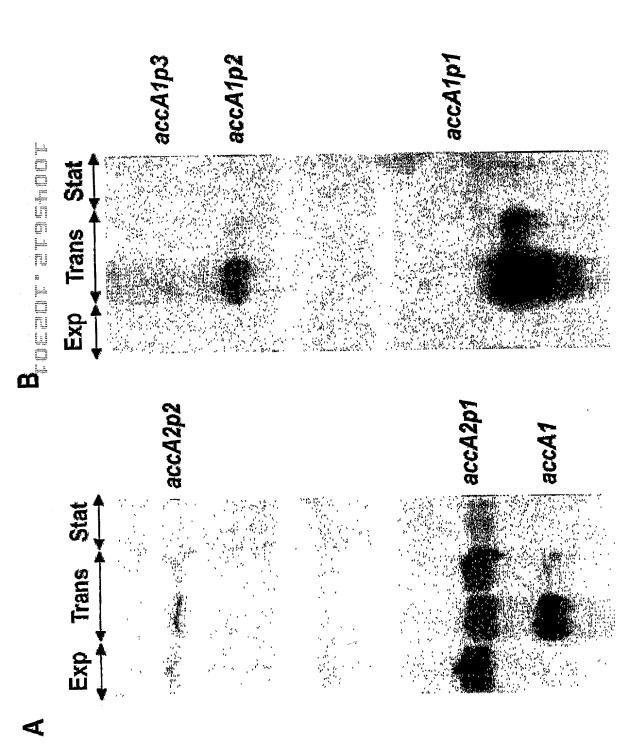
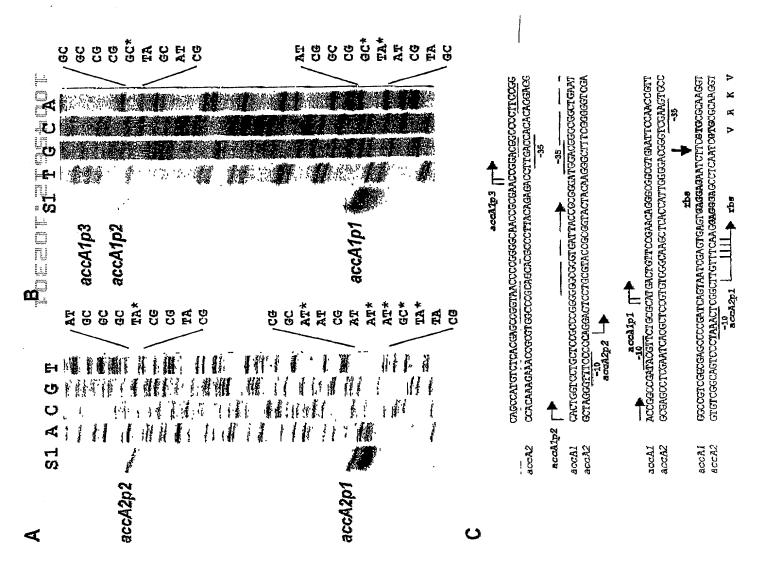
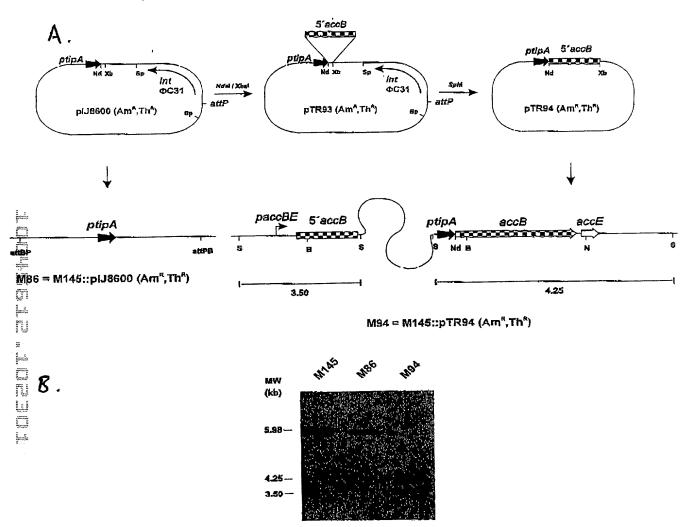


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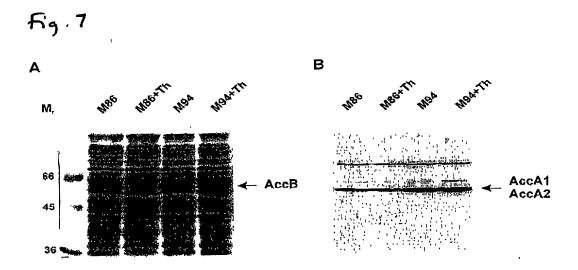




Construction and analysis of the accBE conditional mutant

A. Diagram showing the integration of plus 600 in strain M86 and the expected organization of the Campbell integration of pTR94 in M94. Restriction sites: B, BamHI;N, NotI; Nd, NdeI; S, SacI; Sp, SphI; Xb, XbaI.

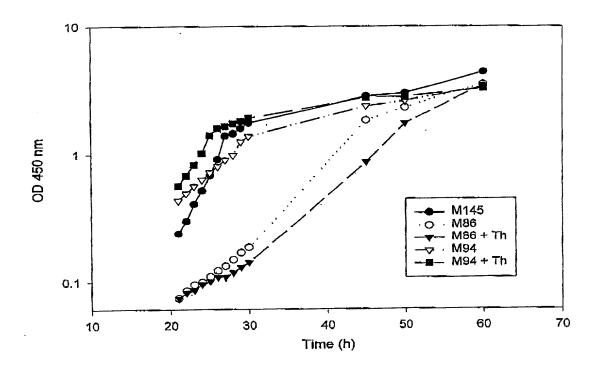
B. Hybridization analysis of Southern blot of SacI-digested DNAs from M145, M86 and M94. The probe was the internal NdeI-XbaI fragment of accB showed in A.



Expression of the acyl-CoA components in M86 and M94

A. SDS-PAGE of cell-free extracts of S. coelicolor M86 and M94 strains grown in YEME medium containing 10 μg ml¹ Am with or without the addition of 5 μg ml¹ Th. B. A duplicate of the SDS-PAGE gel showed in A was subjected to Western blotting and stained for biotinylated proteins by using alkaline phophatase-streptavidin conjugate.

Fig. 8A



Growth curves of M145, M86 and M94 strains. 10^8 spores of strains M86 and M94 were inoculated in YEME medium containing 10 μg of Am or 10 μg ml⁻¹ Am and 5 μg ml⁻¹ of Th. 10^8 spores of M145 were inoculated in YEME. The growth was followed by measuring OD $_{450\,nm}$.

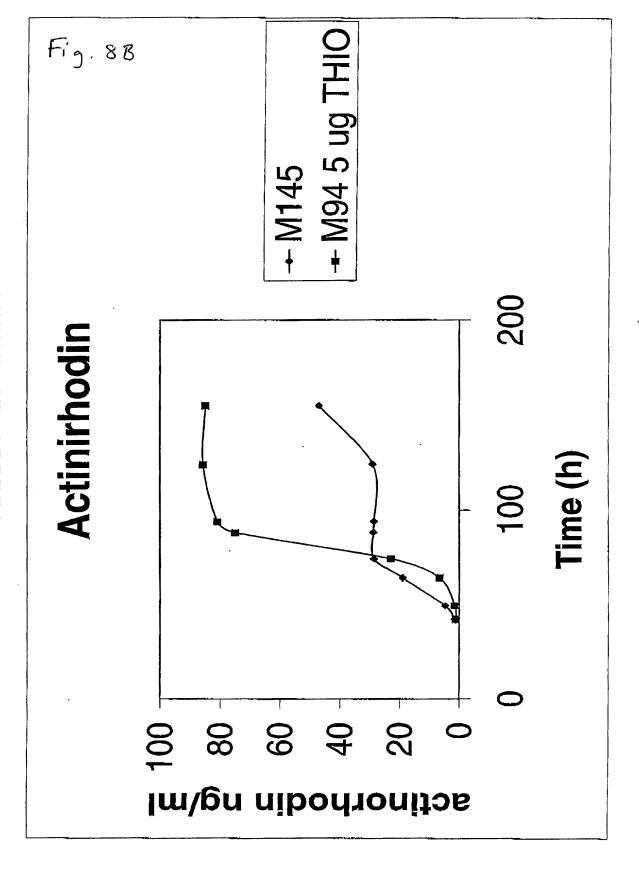
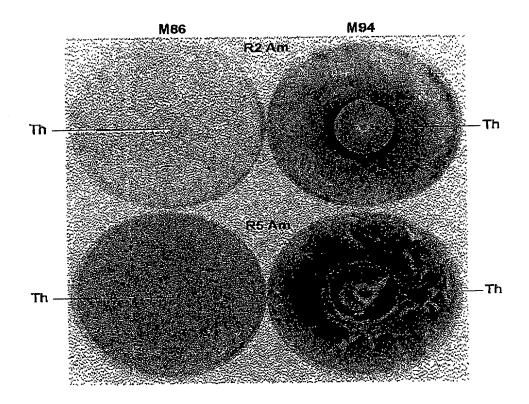


Fig. 9



Morphological and physiological differentiation of M86 and M94 in the precense of Th. Spores of M86 and M94 were spread in R2 or R5 media containing 10 μ g mΓ¹ Am. A drop containing 1 μ g of Th was spotted in the centre of each plate. The picture shows the results obtained after the incubation of the plate at 30 °C for 48 h.

Fig. 11

A. Accal

VRKVLIANRGEIAVRVARACRDAGIASVAVYADPDRDALHVRAADEAFALGGDTPATSYLDIAKVL
KAARESGADAIHPGYGFLSENAEFAQAVLDAGLIWIGPPPHAIRDRGEKVAARHIAQRAGAPLVAG
TPDPVSGADEVVAFAKEHGLPIAIKAAFGGGGRGLKVARTLEEVPELYDSAVREAVAAFGRGECFV
ERYLDKPRHVETQCLADTHGNVVVVSTRDCSLQRRHQKLVEEAPAPFLSEAQTEQLYSSSKAILKE
AGYGGAGTVEFLVGMDGTIFFLEVNTRLQVEHPVTEEVAGIDLVREMFRIADGEELGYDDPALRGH
SFEFRINGEDPGRGFLPAPGTVTLFDAPTGPGVRLDAGVESGSVIGPAWDSLLAKLIVTGRTRAEA
LQRAARALDEFTVEGMATAIPFHRTVVRDPAFAPELTGSTDPFTVHTRWIETEFVNEIKPFTTPAD
TETDEESGRETVVVEVGGKRLEVSLPSSLGMSLARTGLAAGARPKRRAAKKSGPAASGDTLASPMQ
GTIVKIAVEEGQEVQEGDLIVVLEAMKMEQPLNAHRSGTIKGLTAEVGASLTSGAAICEIKD

A. AccA2

VRKVLIANRGEIAVRVARACRDAGIASVAVYADPDRDALHVRAADEAFALGGDTPATSYLDIAKVL KAARESGADAIHPGYGFLSENADFAQAVLDAGLIWIGPPPHAIRDRGEKVAARHIAQRAGAPLVAG TPDPVSGADEVVAFAKEHGLPIAIKAAFGGGGRGLKVARTLEEVPELYDSAVREAVAAFGRGECFV ERYLDKPRHVETQCLADTHGNVVVVSTRDCSLQRRHQKLVEEAPAPFLSEAQTEQLYSSSKAILKE AGYVGAGTVEFLVGMDGTISFLEVNTRLQVEHPVTEEVAGIDLVREMFRIADGEELGYDDPALRGH SFEFRINGDHPGRGFLPAPGTVTLFDAPTGPGVRLDAGVESGSVIGPAWDSLLAKLIVTGRTRAEA LQRAARALDEFTVEGMATAIPFHRTVVRDPAFAPELTGSTDPFTVHTRWIETEFVNEIKPFTTPAD TETDEESGRETVVVEVGGKRLEVSLPSSLGMSLARTGLAAGARPKRRAAKKSGPAASGDTLASPMQ GTIVKIAVEEGQEVQEGDLIVVLEAMKMEQPLNAHRSGTIKGLTAEVGASLTSGAAICEIKD

B. accAl

gtgcgcaaggtgctcatcgccaatcgtggcgaaatcgctgtccgcgtggcccgggcctgccgggac gccgggatcgcgagcgtggccgtctacgcggatccggaccgggacgcgttgcacgtccgtgccgct qatqaqqqqttcqccctqggtggtqacaccccqcgaccagctatctggacatcgccaaggtcctc aaagccgcgcgagtcgggcgcggacgccatccaccccggctacggattcctctcggagaacgcc acccccqaccccgtctccggcgcggacgaggtcgtcgccttcgccaaggagcacggcctgcccatc qccatcaaqqccqccttcgqcgqcgqgqgqqqqcqtcaaggtcgcccqcaccctcgaagaggtg $\verb|ccggagctgtacgactccgcgtgcgcgtggccgccttcggccgcggggagtgcttcgtc|\\$ gagegetacetegacaageceegecacgtggagacecagtgcetggecgacacecaeggcaacgtg gtcgtcgtctccacccgcgactgctccctccagcgccgccaccaaaagctcgtcgaggaggccccc gogocotttototoogaggocoagacggagcagctgtactcatcctccaaggccatcctgaaggag gccggctacggcgccggcaccgtggagttcctcgtcggcatggacggcacgatcttcttcctg gaggtcaacacccgcctccaggtcgagcacccggtcaccgaggaagtcgccggcatcgacttggtc cgcgagatgttccgcatcgccgacggcgaggaactcggttacgacgaccccgccctgcgcggccac inaccetettegaegegeceaeeggeceeggegteegeetggaegeeggegtegagteeggeteeg mtcatcggccccgcctgggactccctcctcgccaaactgatcgtcaccggccgcacccgcgcgagg Lacactocagogogogogogogocotggacgagttoaccgtogagggcatggccacogccatecoct | tocacogcacggtcgtccgcgacccggccttcgccccgaactcaccggctccacggaccccttca acaccgagacggacgaggagtcggggaggacggtcgtcgtcgaggtcggcggcaagcgcctgg __aagtetecetecetecageetgggeatgtecetggeecgeaceggeetggeegeeggggeeegee Ccaagegegegegegaagaagteeggeeeegeetegggegaeaceetegeeteeegatge agggcacgatcgtcaagatcgccgtcgaggaaggccaggaagtccaggaaggcgacctcatcgtcg tactcgaggcgatgaagatggaacagcccctcaacgcccacaggtccggcaccatcaagggcctca ccqccqaqqtcgqcqcctccctcacctccggcgcccatctgcgagatcaaggactga

B. accA2

gtgcgcaaggtgctcatcgccaatcgtggcgaaatcgctgtccgcgtggcccgggcctgccgggac gccgggatcgcgagcgtggccgtctacgcggatccggaccgggacgcgttgcacgtccgtgccgct gatgaggcgttcgccctgggtggtgacaccccgcgaccagctatctggacatcgccaaggtcctc aaagccgcgcgagtcgggcgcggacgccatccaccccggctacggattcctctcggagaacgcc gaccgtggcgaaaaggtcgccgccgccacatcgcccagcgggccggcgcccccctggtcgccgc acccccgaccccgtctccggcgcggacgaggtcgtcgccttcgccaaggagcacggcctgcccatc gccatcaaggccgccttcggcggcgggggggggcctcaaggtcgcccgcaccctcgaagaggtg ccggagctgtacgactccgccgtccgcgaggccgtggccgccttcggccgcggggagtgcttcgtc gagcgctacctcgacaagccccgccacgtggagacccagtgcctggccgacacccacggcaacgtg gtcgtcgtctccacccgcgactgctccctccagcgccgccaccaaaagctcgtcgaggaggccccc gcgccctttctctccgaggcccagacggagcagctgtactcatcctccaaggccatcctgaaggag gccggctacggcggcgccggcaccgtggagttcctcgtcggcatggacggcacgatcttcttcctg _gaggtcaacacccgcctccaggtcgagcacccggtcaccgaggaagtcgccggcatcgacttggtc __cgcgagatgttccgcatcgccgacggcgaggaactcggttacgacgaccccgccctgcgcggccac #accctcttegacgcgcccaccggccccggcgtccgcctggacgccggcgtcgagtccggctccgtc #atcggccccgcctgggaotccctcctcgccaaactgatcgtcaccggccgcacccgcgccgaggca =ctocagegegegegecetggaegagtteacegtegagggcatggecacegecateceette fibaccgcacggtcgtccgcgacccggccttcgccccgaactcaccggctccacggaccccttcacc maagcgccgcgcgccaagaagtccggccccgccgcctcgggcgacaccctcgcctccccgatgcag aggcacgatcgtcaagatcgccgtcgaggaaggccaggaagtccaggaaggcgacctcatcgtcgta etcgaggcgatgaagatggaacagccctcaacgcccacaggtccggcaccatcaagggcctcacc gccgaggtcggcgcctccctcacctccggcgcccatctgcgagatcaaggactga

Fig. 12

A. AccB

MTVLDEAPGEPTDARGRVAELHGIRAAALAGPSEKATAAOHAKGKLTARERIELLLDPGSFREVEO LRRHRATGFGLEAKKPYTDGVITGWGTVEGRTVFVYAHDFRIFGGALGEAHATKIHKIMDMAIAAG APLVSLNDGAGARIQEGVSALAGYGGIFQRNTKASGVIPQISVMLGPCAGGAAYSPALTDFVFMVR DTSQMFITGPDVVKAVTGEEITQNGLGGADVHAETSGVCHFAYDDEETCLAEVRYLLSLLPQNNRE NPPRAESSDPVDRRSDTLLDLVPADGNRPYDMTKVIEELVDEGEYLEVHERWARN1ICALARLDGR VVGIVANQPQALAGVLDIEASEKAARFVQMCDAFNIPIITLLDVPGFLPGVDQEHGGIIRHGAKLL YAYCNATVPRISLILRKAYGGAYIVMDSQSIGADLTYAWPTNEIAVMGAEGAANVIFRRQIADAED PEAMRARMVKEYKSELMHPYYAAERGLVDDVIDPAETREVLITSLAMLHTKHADLPSRKHGNPPO

B. accB

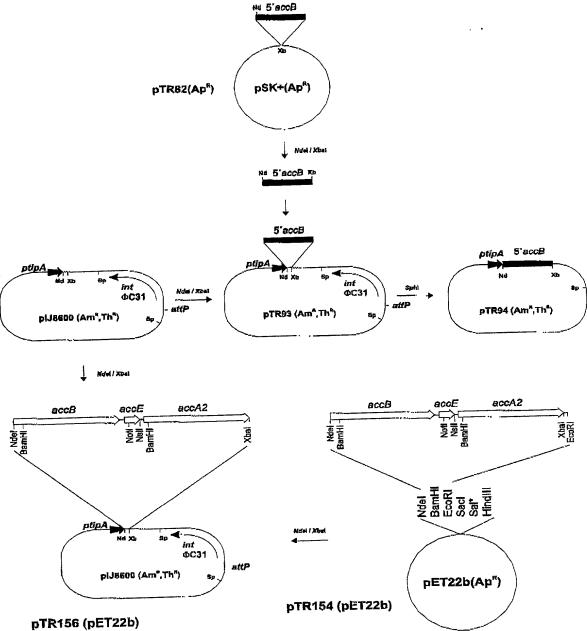
AAGCTGACGCACGTGAGCGCATCGAGCTCCTCGGACCCCGGCTCCTTCCGCGAGGTCGAGCAG CTCCGCCGCCACCGGCGACCGGGTTCGGCCTGGAGGCCCAAGAAGCCGTACACCGACGGTGTCATC ACCGCTGGGCACGGTCGAGGGCCGCACGGTCTTCGTCTACGCCCACGACTTCCGGATCTTCGGC GGCGCGCTGGGCGAGGCCCACGCCACGAAGATCCACAAGATCATGGACATGGCCATCGCGGCCGGT GCCCGCTGGTGTCGCTGAACGACGCCCCGCCCCGTATCCAGGAGGGCGTCAGCGCCCTCGCC GGGTACGCCGCATCTTCCAGCGCAACACCAAGGCGTCCGGCGTCATCCCGCAGATCAGCGTGATG $\verb|CTCGGCCCTGCGGGGGGGGGGGCGTACAGCCCCGCCTTACCGACTTCGTCTTCATGGTCCGC||$ GACACCTCGCAGATGTTCATCACGGGCCCGGACGTCGTCAAGGCGGTCACCGGCGAGGAGATCACG CAGAACGGTCTGGGCGCGCCGACGTGCACGCCGAGACGTCCGGCGTGTGCCACTTCGCCTACGAC AACCGCCCCGCGCGAGTCCTCCGACCCCGTGGACCGCCGCTCGGACACCCTCCTCGACCTGGTC CCGCCGACGCCAACCGCCCGTACGACATGACCAAGGTCATCGAGGAACTCGTCGACGAGGGCGAG TACCTGGAGGTCCACGAGCGTTGGGCCCGCAACATCATCTGCGCGCCTGGCCCGTCTCGACGGGCGG GTCGTGGGCATCGTCGCCAACCAGCCGCAGGCCCTGGCCGGTGTCCTGGACATCGAGGCGTCGGAG AAGGCGGCCCGCTTCGTCCAGATGTGCGACGCCTTCAACATCCCGATCATCACTCTTCTGGACGTA CCCGCCTTCCTGCCCGCGTCGACCAGGAGCACGCGGGGATCATCCGCCACGGCGCCAAGCTGCTC TACGCGTACTGCAACGCGACCGTGCCCCGGATCTCGCTGATCCTGCGCAAGGCGTACGGAGGTGCT TACATCGTCATGGACAGCCAGTCCATCGGCGCCGACCTCACCTACGCCTGGCCGACCAACGAGATC GCCGTCATGGGCGCGAAGGTGCCGCGAACGTCATCTTCCGCCGGCAGATCGCCGACGCCGAGGAC CCCGAGGCCATGCGGGCGCGCATGGTCAAGGAGTACAAGTCCGAGCTGATGCACCCCTACTACGCG GCCGAACGCGGTCTGGTCGACGACGTCATCGACCCCGCCGAAACCCGCGAGGTGCTGATCACGTCC CTGGCGATGCTCCACACCAAGCACGCCGACCTGCCCTCCCGCAAGCACGGCAACCCGCCGCAGTGA Fig. 13

A. AccE

MSPADIRVEKGHAEPEEVAAITALLLARAAARPAEIAPTHGGGRARAGWRRLEREPGFRAPHSWR

B. accE

Fig. 14



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